Short Communication

Diverse responses of belowground internal nitrogen cycling to increasing aridity

Dan Kou[a,b], Yunfeng Peng[a], Guanqin Wang[a,b], Jinzhi Ding[a,b], Yongliang Chen[a], Guibiao Yang[a,b], Kai Fang[a,b], Li Liu[a,b], Beibei Zhang[a], Christoph Müller[c,d], Jinbo Zhang[e,**], Yuanhe Yang[a,b,*]

[a] State Key Laboratory of Vegetation and Environmental Change, Institute of Botany, Chinese Academy of Sciences, Beijing 100093, China
[b] University of Chinese Academy of Sciences, Beijing 100049, China
[c] Department of Plant Ecology, Justus-Liebig University Giessen, Heinrich-Buff-Ring 26, 35392 Giessen, Germany
[d] School of Biology and Environmental Science, University College Dublin, Ireland
[e] School of Geography Sciences, Nanjing Normal University, Nanjing 210047, China

ARTICLE INFO

ABSTRACT

Belowground microbial nitrogen (N) processes play key roles in regulating terrestrial ecosystem services such as vegetation production, however, our understanding of their responses to climate change remains limited. We determined patterns and controls of five gross N transformation processes along a typical aridity gradient on the Tibetan Plateau. Potential gross N transformation rates responded diversely to the changing aridity. Both mineralization (M_N, average rate: 2.87 mg N kg^{-1} soil d^{-1}) and ammonium immobilization (I_{NH4}, 3.35 mg N kg^{-1} soil d^{-1}) declined as aridity increased. Autotrophic nitrification (O_{NH4}, 1.72 mg N kg^{-1} soil d^{-1}) exhibited a bell-shaped pattern along the gradient, with an optimum aridity of 0.53 (1- aridity index (AI)). By contrast, rates of nitrate immobilization (I_{NO3}, 0.46 mg N kg^{-1} soil d^{-1}) and dissimilatory nitrate reduction to ammonium ($DNRA$, 0.10 mg N kg^{-1} soil d^{-1}) did not respond to the changing aridity. These results suggest that predicted increases in aridity will exert different effects on various soil internal N cycling processes, and thus potentially have profound impact on structure and function of dryland ecosystems.

Drylands, the largest terrestrial biome and one of the most sensitive ecosystems to environmental changes, provide crucial ecosystem services (*e.g.*, food and fibre production, carbon sequestration) for more than 38% of the global population (Maestre et al., 2012; Schimel, 2010). Although primarily limited by water availability, ecosystem services in drylands are secondarily limited by nitrogen (N) availability (Delgado-Baquerizo et al., 2016). Therefore, ecosystem N cycling as a function of aridity, the major climate driver in drylands, has attracted increasing interest. However, previous studies focused mainly on the effect of changing aridity on state variables related to N cycling (*e.g.*, total N (TN), available N and δ^{15}N; Delgado-Baquerizo et al., 2013a, 2016; Wang et al., 2014), with notably less attention paid on belowground gross N transformations. Given that biogeochemical N cycling and the availability of N in the biosphere are governed by a series of simultaneously occurring N transformation processes (Booth et al., 2005; Chapin et al., 2011), it is essential to reveal patterns and controls of belowground microbial N processes along aridity gradients in drylands.

To examine the potential responses of soil microbial N processes to aridity, we collected surface soil (0-10 cm) samples from 18 sites along an aridity gradient on the Tibetan Plateau during the summer (July and August) of 2014 (Table S1). With those soil samples, we quantified a set of simultaneously occurring gross N transformation rates using a ^{15}N-tracing technique and also determined a suite of physical and chemical properties and microbial characteristics, including phospholipid fatty acids (PLFA) and *amoA* gene abundance of ammonia oxidizing archaea (AOA) and bacteria (AOB). In addition, the aridity index (AI, the ratio of precipitation to potential evapotranspiration) was extracted from the CGIAR-CSI Global-Aridity and Global-PET database (http://www.cgiar-csi.org) (Zorner et al., 2008). The relative importance of biotic and abiotic variable on gross N transformation rates was assessed using a multi-model inference approach (Maestre et al., 2012). See more details in the supplementary materials.

Our results showed that gross N transformation rates, including gross N mineralization (M_N), ammonium (NH$_4^+$) immobilization (I_{NH4}), autotrophic nitrification (O_{NH4}), nitrate (NO$_3^-$) immobilization (I_{NO3}) and dissimilatory nitrate reduction to ammonium ($DNRA$), exhibited large spatial variability on the Tibetan Plateau (Fig. 1 and Fig. S1).
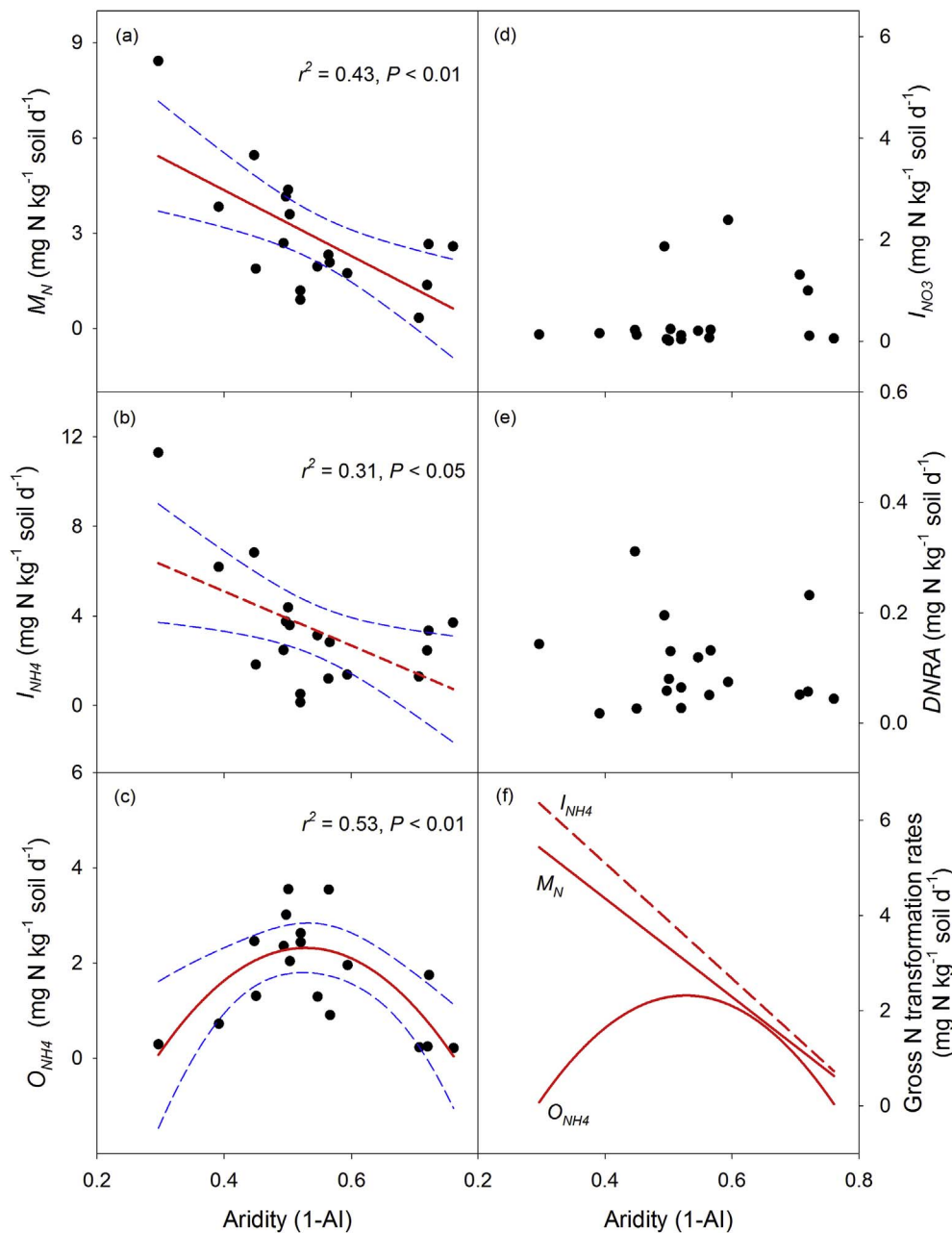
Fig. 1. Variations of soil microbial gross N transformation rates along a typical aridity gradient on the Tibetan Plateau. a, M_N, gross N mineralization; b, I_{NH4}, immobilization of NH_4^+; c, O_{NH4}, autotrophic nitrification; I_{NO3}, immobilization of NO_3^-; d, DNRA, dissimilatory NO_3^- reduction to NH_4^+. To facilitate the explanation of our results, aridity is defined as 1-AI, where AI (aridity index) is the ratio of precipitation to evapotranspiration. The red lines and blue dashed lines represent the fitted curves and their 95% confidence intervals, respectively. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Among these rates, both M_N and I_{NH4} were negatively correlated with aridity (1-AI) (M_N: $P < 0.01$; I_{NH4}: $P < 0.05$; Fig. 1a-b). Consistent with the decrease of M_N and I_{NH4} along the aridity gradient, our results revealed that factors that tend to promote the rates of M_N and I_{NH4}, such as plant cover, clay content, soil organic matter, available substrate (dissolved organic N (DON) for M_N; NH_4^+ for I_{NH4}) and total microbial biomass, declined with increasing aridity, whereas variables such as pH that tend to inhibit M_N and I_{NH4} increased with aggravated drought (Figs. S2-3, S5). These results suggested that the interaction of vegetation, soil environment, substrate and microbial community along the aridity gradient caused the decreases in rates of M_N and I_{NH4} with the increasing aridity. Among all factors, available substrate was the most important in regulating the variations in rates of both M_N and I_{NH4} (Fig. 2a-b, Table S2). Moreover, neither M_N nor I_{NH4} exhibited any significant relationships with soil SOC:TN, F:B or G+:G-ratio (Figs. S2–3), suggesting that substrate quality and microbial community composition were not the significant drivers for M_N and I_{NH4} along this aridity gradient.

Autotrophic nitrification exhibited a bell-shaped pattern along the aridity gradient, with the maximum rate occurring at aridity (1-AI) = 0.53 ($P < 0.01$; Fig. 1c). Such a relationship could be explained by the similar pattern of ammonia supplying capacity (reflected by M_N - I_{NH4}, the difference between M_N and I_{NH4}) and the abundance of AOA along the aridity gradient (Fig. S5k-l), which was further demonstrated by the significant linear association between O_{NH4} and M_N - I_{NH4} or AOA (Figs. S4g–h) and the large importance values of M_N - I_{NH4} and AOA (Fig. 2c, Table S2). A question then is why M_N - I_{NH4} and AOA exhibited bell-shaped patterns along the aridity gradient. Before approaching the optimum aridity (aridity (1-AI) = 0.53, i.e., the relatively wet region), we expected that the increase in ammonia supplying capacity and AOA abundance that occurred with increasing aridity could be largely due to decreased competition of heterotrophic microorganisms as well as plants for ammonium against nitrifiers (Chapin et al., 2011; Delgado-Baquerizo et al., 2013b). However, the decline in ammonia supplying capacity and AOA abundance after reaching the optimum aridity (aridity (1-AI) = 0.53, i.e., the relatively dry region) might be attributed to
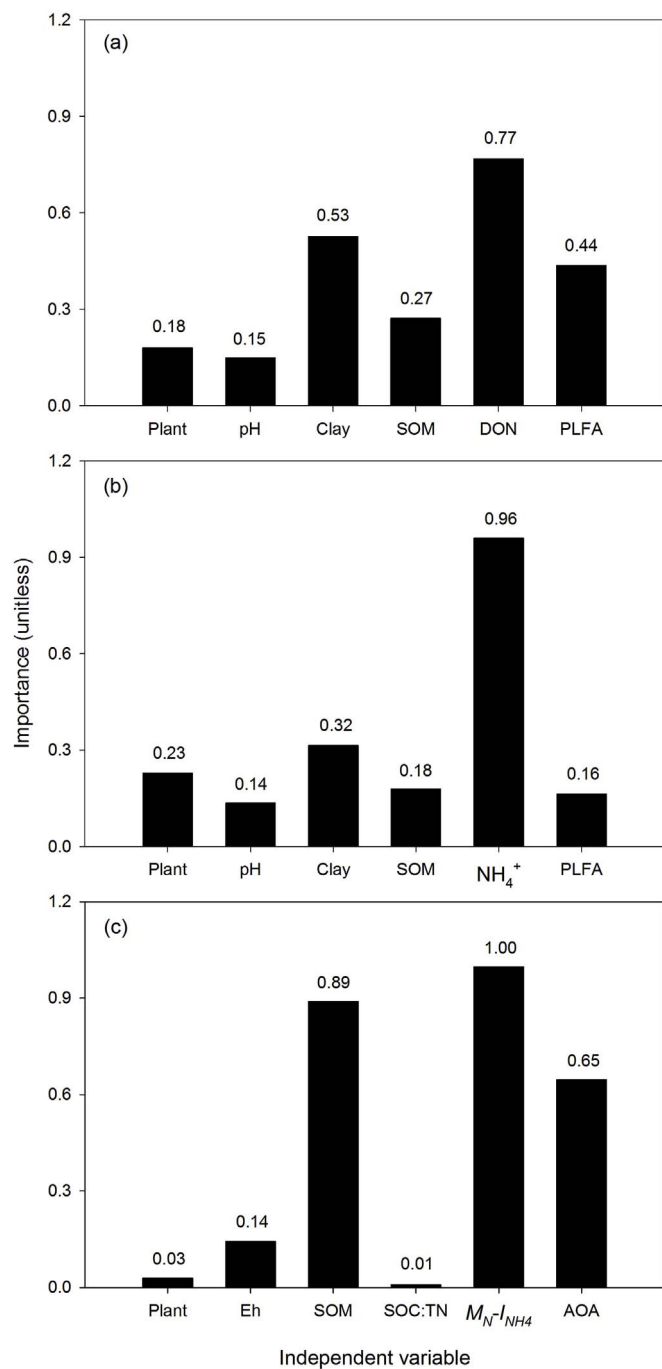
Fig. 2. Relative importance of vegetation (plant cover), edaphic environment (pH, clay and Eh), substrate (SOM, DON, NH_4^+, SOC:TN, M_N-I_{NH4}) and microbial properties (total PLFAs and AOA) as predictors of gross N mineralization rate (M_N, a), NH_4^+ immobilization rate (I_{NH4}, b) and autotrophic nitrification rate (O_{NH4}, c) in drylands. The importance score of each bar is the sum of the Akaike weights of all models (63 models for each response variable) that include the predictor of interest, analyzed by a multi-model inference approach. Eh indicates soil redox potential. SOM reflects soil organic matter, which is the first component of a principal component analysis (PCA) conducted with soil organic carbon (SOC) and total nitrogen (TN) and explains 99.2% of the total variance. DON represents dissolved organic nitrogen. M_N - I_{NH4} is the difference between M_N and I_{NH4}, which is used to reflect the ammonia supplying capacity for nitrification. Total PLFAs is used to reflect the total microbial biomass in soils. AOA is ammonia oxidizing archaea.

the decrease in N mineralization rate as soil organic matter content was reduced and drought stress aggravated (Booth et al., 2005; Delgado-Baquerizo et al., 2013b).

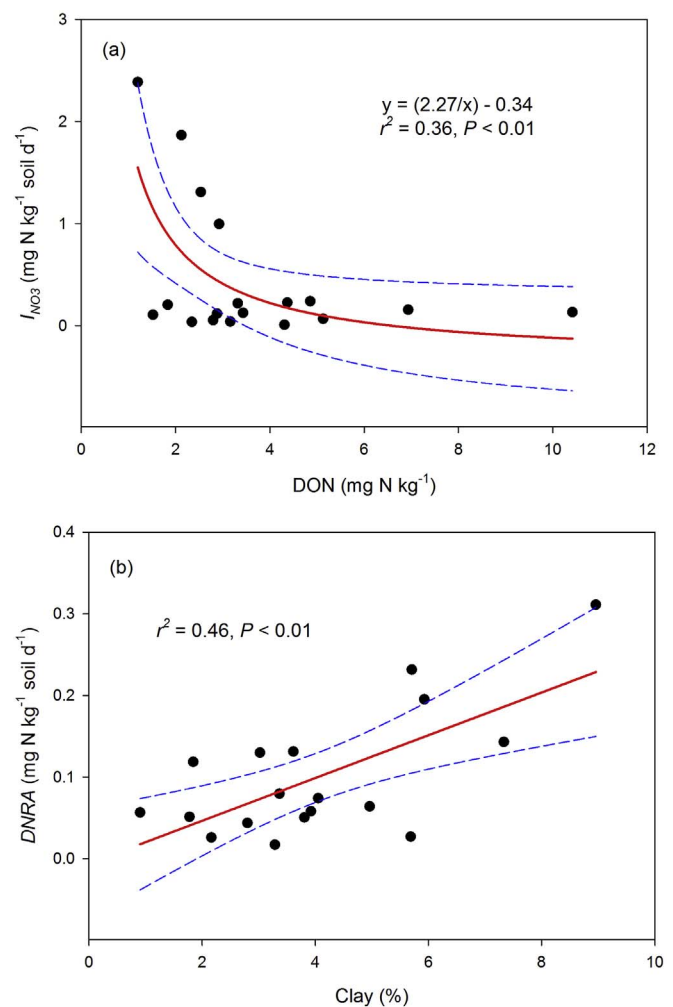Although the transformations associated with the turnover of NH_4^+

Fig. 3. Relationships of NO_3^- immobilization rate (I_{NO3}) with DON (a) and dissimilatory NO_3^- reduction to NH_4^+ rate ($DNRA$) with clay content (b). The red lines and blue dashed lines represent the fitted curves and their 95% confidence intervals, respectively. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

showed significant relationships with aridity, this was not the case for the N transformations associated with NO_3^- turnover (I_{NO3} and $DNRA$; Fig. 1d-e). Both I_{NO3} and $DNRA$ were less responsive to aridity, most likely because of the low precipitation and low level of N leaching in drylands, although significant relationships of I_{NO3} and $DNRA$ with aridity index have been frequently observed in forest ecosystems with high precipitation and levels of leaching (Huygens et al., 2008; Rütting et al., 2011; Zhang et al., 2011). Our results showed that I_{NO3} in drylands was primarily controlled by soil DON availability (Fig. 3a). The rapid increase in I_{NO3} with the decrease of DON indicated that microbes assimilated NO_3^- when DON was not freely available in soil to meet microbial requirements for N. Moreover, we found that $DNRA$ was positively associated with clay content (Fig. 3b), possibly reflecting that high clay content can generate a relatively reductive environment, which could provide suitable conditions for $DNRA$ (Paul, 2006; Sotta et al., 2008).

Collectively, our results demonstrated that simultaneously-occurring soil microbial N cycling processes responded diversely to increasing aridity, with linear decreases in mineralization and NH_4^+ immobilization, bell-shaped pattern in nitrification, and no responses in NO_3^- immobilization and $DNRA$ along the aridity gradient. Given that projections are that aridity will increase in many regions worldwide (Delgado-Baquerizo et al., 2013a), the patterns observed in this study suggest that these changes will exert differential effects on different soil

internal N cycling processes. Consequently, increased aridity may alter plant N availability and thus have profound impact on structure and function of dryland ecosystems.

Acknowledgments

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.soilbio.2017.10.010.

References

Booth, M.S., Stark, J.M., Rastetter, E., 2005. Controls on nitrogen cycling in terrestrial ecosystems: a synthetic analysis of literature data. Ecological Monographs 75, 139–157.

Chapin, F.S., Matson, P.A., Vitousek, P., 2011. Principles of Terrestrial Ecosystem Ecology. Springer, New York.

Delgado-Baquerizo, M., Maestre, F.T., Gallardo, A., Bowker, M.A., Wallenstein, M.D., Quero, J.L., Ochoa, V., Gozalo, B., Garcia-Gomez, M., Soliveres, S., Garcia-Palacios, P., Berdugo, M., Valencia, E., Escolar, C., Arredondo, T., Barraza-Zepeda, C., Bran, D., Carreira, J.A., Chaieb, M., Conceicao, A.A., Derak, M., Eldridge, D.J., Escudero, A., Espinosa, C.I., Gaitan, J., Gatica, M.G., Gomez-Gonzalez, S., Guzman, E., Gutierrez, J.R., Florentino, A., Hepper, E., Hernandez, R.M., Huber-Sannwald, E., Jankju, M., Liu, J., Mau, R.L., Miriti, M., Monerris, J., Naseri, K., Noumi, Z., Polo, V., Prina, A., Pucheta, E., Ramirez, E., Ramirez-Collantes, D.A., Romao, R., Tighe, M., Torres, D., Torres-Diaz, C., Ungar, E.D., Val, J., Wamiti, W., Wang, D., Zaady, E., 2013a. Decoupling of soil nutrient cycles as a function of aridity in global drylands. Nature 502, 672–676.

Delgado-Baquerizo, M., Gallardo, A., Wallenstein, M.D., Maestre, F.T., 2013b. Vascular plants mediate the effects of aridity and soil properties on ammonia-oxidizing bacteria and archaea. FEMS Microbiology Ecology 85, 273–282.

Delgado-Baquerizo, M., Maestre, F.T., Gallardo, A., Eldridge, D.J., Soliveres, S., Bowker, M.A., Prado-Comesaña, A., Gaitán, J., Quero, J.L., Ochoa, V., Gozalo, B., García-Gómez, M., García-Palacios, P., Berdugo, M., Valencia, E., Escolar, C., Arredondo, T., Barraza-Zepeda, C., Boeken, B.R., Bran, D., Cabrera, O., Carreira, J.A., Chaieb, M., Conceição, A.A., Derak, M., Ernst, R., Espinosa, C.I., Florentino, A., Gatica, G., Ghiloufi, W., Gómez-González, S., Gutiérrez, J.R., Hernández, R.M., Huber-Sannwald, E., Jankju, M., Mau, R.L., Miriti, M., Monerris, J., Morici, E., Muchane, M., Naseri, K., Pucheta, E., Ramírez, E., Ramírez-Collantes, D.A., Romão, R.L., Tighe, M., Torres, D., Torres-Díaz, C., Val, J., Veiga, J.P., Wang, D., Yuan, X., Zaady, E., 2016. Human impacts and aridity differentially alter soil N availability in drylands worldwide. Global Ecology and Biogeography 25, 36–45.

Huygens, D., Boeckx, P., Templer, P., Paulino, L., Van Cleemput, O., Oyarzún, C., Müller, C., Godoy, R., 2008. Mechanisms for retention of bioavailable nitrogen in volcanic rainforest soils. Nature Geoscience 1, 543–548.

Maestre, F.T., Quero, J.L., Gotelli, N.J., Escudero, A., Ochoa, V., Delgado-Baquerizo, M., Garcia-Gomez, M., Bowker, M.A., Soliveres, S., Escolar, C., Garcia-Palacios, P., Berdugo, M., Valencia, E., Gozalo, B., Gallardo, A., Aguilera, L., Arredondo, T., Blones, J., Boeken, B., Bran, D., Conceicao, A.A., Cabrera, O., Chaieb, M., Derak, M., Eldridge, D.J., Espinosa, C.I., Florentino, A., Gaitan, J., Gatica, M.G., Ghiloufi, W., Gomez-Gonzalez, S., Gutierrez, J.R., Hernandez, R.M., Huang, X., Huber-Sannwald, E., Jankju, M., Miriti, M., Monerris, J., Mau, R.L., Morici, E., Naseri, K., Ospina, A., Polo, V., Prina, A., Pucheta, E., Ramirez-Collantes, D.A., Romao, R., Tighe, M., Torres-Diaz, C., Val, J., Veiga, J.P., Wang, D., Zaady, E., 2012. Plant species richness and ecosystem multifunctionality in global drylands. Science 335, 214–218.

Paul, E.A., 2006. Soil Microbiology, Ecology and Biochemistry. Academic press, New York.

Rütting, T., Boeckx, P., Müller, C., Klemedtsson, L., 2011. Assessment of the importance of dissimilatory nitrate reduction to ammonium for the terrestrial nitrogen cycle. Biogeosciences 8, 1779–1791.

Schimel, D.S., 2010. Drylands in the earth system. Science 327, 418–419.

Sotta, E.D., Corre, M.D., Veldkamp, E., 2008. Differing N status and N retention processes of soils under old-growth lowland forest in Eastern Amazonia, Caxiuanã, Brazil. Soil Biology and Biochemistry 40, 740–750.

Wang, C., Wang, X.B., Liu, D.W., Wu, H.H., Lu, X.T., Fang, Y.T., Cheng, W.X., Luo, W.T., Jiang, P., Shi, J.S., Yin, H.Q., Zhou, J.Z., Han, X.G., Bai, E., 2014. Aridity threshold in controlling ecosystem nitrogen cycling in arid and semi-arid grasslands. Nature Communications 5. http://dx.doi.org/10.1038/ncomms5799.

Zhang, J., Zhu, T., Cai, Z., Müller, C., 2011. Nitrogen cycling in forest soils across climate gradients in Eastern China. Plant and Soil 342, 419–432.

Zorner, R.J., Trabucco, A., Bossio, D.A., Verchot, L.V., 2008. Climate change mitigation: a spatial analysis of global land suitability for clean development mechanism afforestation and reforestation. Agriculture Ecosystems and Environment 126, 67–80.